5-Fluorocytosine and Urinary Candidiasis

R. R. DAVIES, D. S. REEVES

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Summary

Two patients with urinary candidiasis were treated with 5-fluorocytosine. Serum levels of the drug and a study of its excretion in urine are recorded together with a brief account of its activity in body fluids. Evidence of toxicity was not observed with dosages of from 2 to 5 g a day.

Introduction

Urinary infections with *Candida albicans* are not common (Guze and Haley, 1958; Seneca and Peer, 1965) but can be difficult to eradicate, especially in the presence of a physiological or anatomical abnormality. Until recently the only available systemic antifungal agent was amphotericin B, which is only poorly excreted in the urine (Louria, 1958). Local therapy by irrigation with amphotericin B has been used (Goldman *et al.*, 1960; Blum, 1966) but may not be successful and requires the presence of indwelling ureteric catheters or ureterostomies to ensure treatment of the upper part of the tract.

5-Fluorocytosine is an antimetabolite of the pyrimidine base cytosine showing a high degree of activity against *C. albicans* (Grunberg *et al.*, 1964, 1967) but with relatively little reported toxicity in man (Koechlin *et al.*, 1966; Grunberg *et al.*, 1967; Tassel and Madoff, 1968; Utz *et al.*, 1968). Pharmacological studies have shown that, in man, it is not detectably metabolized and is mostly excreted in the urine in an unchanged form (Koechlin *et al.*, 1966). This suggests that high urinary levels of 5-fluorocytosine may be obtained with moderate dosage, even when some degree of renal failure is present, and that 5-fluorocytosine may therefore be useful for treating candida urinary infections.

In this paper we describe the treatment of two patients with urinary candidiasis and record an investigation of the pharmacology of 5-fluorocytosine, including laboratory studies on its activity in body fluids.

Clinical Observations

CASE 1

The patient, a married woman, aged 66 (weight 50 kg), had had chronic bladder outlet obstruction following the removal of a urethral caruncle 18 years previously. She had developed gross bilateral hydronephrosis and hydroureters with widely dilated urethral orifices. The bladder was trabeculated and showed multiple wide-mouth saccules. On admission with a further episode of retention her urine was found to be infected with a *Staphylococcus aureus* (10^5 /ml) and *C. albicans* (10^4 /ml). The presence of the fungus infection was confirmed in urine taken by suprapubic bladder puncture. The staphylococcal infection was eradicated by a course of cephaloridine but the candida infection persisted.

Department of Bacteriology, St. Mary's Hospital Medical School, London W.2

R. R. DAVIES, M.Sc., Ph.D., Senior Lecturer

D. S. REEVES, M.B., M.R.C.PATH., Lecturer. (Present appointment: Consultant Bacteriologist, Southmead Hospital, Bristol BS10 5NB)

The presence of free ureteric reflux made upper urinary tract infection, with the risks of renal abscess formation and systemic infection, a strong possibility (Davis *et al.*, 1956). It was therefore considered necessary to give systemic therapy. A pre-existing impairment of renal function excluded the use of amphotericin B so 5-fluorocytosine was considered to be appropriate treatment. Treatment with 5-fluorocytosine was therefore given, initially in a dose of 2.5 g daily in divided doses for eight days but subsequently 5 g daily for a further 12 days. Twenty-four hour specimens of urine were collected for the estimation of 5-fluorocytosine concentration and excretion, and blood samples were taken for the estimation of 5-fluorocytosine serum levels.

The catheter was removed after nine days of treatment with 5-fluorocytosine but had to be reinserted for a further period when the patient developed another episode of urinary retention accompanied by a bacterial urinary infection and nitrogen retention; this infection was eradicated with a seven-day course of kanamycin. Two days later the patient developed a rash and joint pains which were attributed to one of the antimicrobial agents she had received. Shortly after this she also complained of some deafness, and the audiographic findings suggested this was due to an ototoxic drug, presumably the kanamycin.

Serum levels of 5-fluorocytosine after a single dose are shown in Fig. 1, and the average levels during continuous therapy are shown in Fig. 2, which also shows urinary concentration and total excretion of the drug. The haematological and biochemical results obtained by standard routine methods are shown in Table I.

FIG. 1—Case 1. Serum levels of 5-fluorocytosine at hourly intervals after the first dose as determined in three separate microbiological assays.

TABLE I—Haematological and Biochemical Results in Case 1

Day after Starting Therapy	Haemoglobin (g/100 ml)	White Blood Cells/mm ³		Platelets per mm ³	Blood Urea (g/100 ml)	Comments
		Total	Poly Neut.			
0	11.1	7,000	4,900	446,000	39	Creatinine clearance 31 ml/min
1	10.3	7,000	3,900	439,000	—	Creatinine clearance 50 ml/min
5	11.4	7,000	3,900	435,000	—	
9	—	—	—	—	—	Catheter removed
13	—	—	—	—	—	Kanamycin started
14	—	—	—	—	—	Catheter replaced
18	10.7	7,000	4,200	—	150	End of 5-FC therapy. End of kanamycin therapy
20	—	—	—	—	100	
45	—	—	—	—	74

FIG. 2—Case 1. Serum levels of 5-fluorocytosine, urinary concentrations, urinary output, and total secretion of the drug during continuous therapy.

There was no significant change in the haemoglobin content or white cell count of the peripheral blood. The rise in blood urea was probably associated with the episode of acute retention of urine following the removal of the catheter. It is unlikely to be due to a toxic effect of therapy because the blood urea fell while the drug was continued.

The patient was discharged with a sterile urine six weeks after admission. Subsequent specimens of urine taken at intervals of up to nine months have shown no recurrence of the candida infection, though she has had further episodes of bacterial infection.

CASE 2

This patient, a man, aged 59 (weight 73 kg), was referred with a heavily fibrosed and inoperable abdominal aortic aneurysm causing bilateral ureteric obstruction and uraemia and a history of operative removal of left-sided urinary calculi. Several ureteric catheterizations produced only temporary relief of obstruction and his blood urea rose further. At this stage urine from his urethral and ureteric catheters was found to contain *C. albicans*, about 1×10^5 /ml, in addition to being infected with *Pseudomonas aeruginosa*. Since an obstructive lesion due to candida was suspected the pelvis of the right kidney was explored and found to contain a caseous material which yielded a profuse growth of *C. albicans* and *Escherichia coli*. The serum contained precipitins to an antigen prepared from *C. albicans* cell sap.

Treatment with 5-fluorocytosine (4 g daily) was begun and continued for 10 days. Though the urinary counts of candida were greatly reduced to 2×10^2 /ml the infection persisted, and the drug was stopped when the patient was found to be slightly neutropenic (Fig. 3). His white blood count later returned to normal and there was no evidence of thrombocytopenia on examination of blood smears. Blood and urine samples were taken for the estimation of 5-fluorocytosine and the results are shown in Table II.

The patient's renal function failed to recover and he eventually died four weeks after the course of 5-fluorocytosine finished. Necropsy was not undertaken.

Laboratory Investigations

Assay Procedure.—Assays were performed in glass assay trays 25 by 25 cm, the medium recommended by Shaddomy (1969) and the first patient's strain of *C. albicans* being used.

FIG. 3—Case 2. White blood cell counts.

TABLE II—Case 2. Serum and Urinary Levels of 5-fluorocytosine showing Persistence of Drug after Therapy

Day	Urine		Serum (µg/ml)
	Conc. (µg/ml)	Vol. (ml)	
Treatment			
1	..	..	<0.5
2	..	..	38
3	..	..	108
4	..	..	130
5	..	..	
6	..	..	
7	..	..	
8	..	..	
9	..	..	
10	..	..	
11	..	..	
Treatment ended	..	..	
12	..	..	
13	..	..	75
14	..	..	165
15	..	..	63

Zone diameters for 5-fluorocytosine in serum were found to be the same as those in water. Results were read from a standard curve drawn by plotting log 10 concentrations against zone diameters on a linear scale.

Activity of 5-fluorocytosine in Body Fluids.—Serial doubling dilutions of 5-fluorocytosine in half-strength Wickerham's (1951) medium and Wickerham's medium similarly diluted with sterile urine or serum were inoculated from suspensions of *C. albicans* or *Torulopsis glabrata*. *T. glabrata* was included as a test organism because unlike *C. albicans* it does not produce mycelium in the presence of serum. Yeast cells clump and islets of growth occur in fluids containing 50% human serum and as a consequence growth cannot be reliably estimated by either visual inspection or turbidometrically. All cultures were treated with ultrasonic vibration before making viable counts, and the results of these experiments are given in Tables III and IV.

Serum Binding of 5-fluorocytosine.—A concentration of 100 µg/ml was prepared in serum without significantly diluting the proteins. After equilibration at 37°C for two hours a 5 ml sample was centrifuged at 40,000 r.p.m. (mean g 125,000, temperature about 4°C) for 21 hours. One millilitre of the supernatant was taken for comparison with a "100% free" standard solution of 5-fluorocytosine in buffer. In two separate assays protein binding of 5-fluorocytosine was calculated to be 48 and 49%.

TABLE III—Inactivation of 5-fluorocytosine by Urine

Medium	<i>C. albicans</i> Inoculum	Concentration of 5-fluorocytosine in µg/ml											
		0.5	1	2	4	8	16	32	64	125	250	500	1,000
Wickerham	10 ⁴	+	+	+	+	+	+	+	+	+	+	+	+
	10 ³	+	+	+	+	+	+	+	+	+	+	+	+
	10 ¹	○	○	○	○	○	○	○	○	○	○	○	○
	<10	○	○	○	○	○	○	○	○	○	○	○	○
	10 ⁴	+	+	+	+	+	+	+	+	+	+	+	+
Wickerham diluted to 50% with urine	10 ³	+	+	+	+	+	+	+	+	+	+	+	+
	10 ¹	+	+	+	+	+	+	+	+	+	+	+	+
	<10	+	+	+	○*	+	+	+	+	+	○*	+	○*

+ = Growth. ○ = No growth. *Absence of growth presumed to be due to failure in inoculation when using low-density cell suspensions.

TABLE IV—Enhancement of 5-fluorocytosine Activity in Serum

Yeast	Medium	Concentration of 5-fluorocytosine in µg/ml							
		0.03	0.06	0.125	0.25	0.5	1	2	64
<i>T. glabrata</i>	½-strength Wickerham fluid ..	3.7 × 10 ⁸						2.35 × 10 ⁸	2.4 × 10 ⁸
Mean inoculum 174 cells ..	50% Serum Wickerham fluid ..	4.5 × 10 ⁸	1.8 × 10 ³	9 × 10 ³	125	25	0	—	—
<i>C. albicans</i>	½-strength Wickerham fluid ..	2.8 × 10 ⁸	1.8 × 10 ³	2.4 × 10 ⁷	1.2 × 10 ⁷	4.6 × 10 ⁴	3.7 × 10 ⁸	1 × 10 ⁸	—
Mean inoculum 230 cells ..	50% Serum Wickerham fluid ..	1.8 × 10 ⁸	1.5 × 10 ³	8.5 × 10 ³	9.8 × 10 ³	N.T.	1.2 × 10 ⁸	1.2 × 10 ⁸	—

N.T. = Not tested.

Discussion

In these two patients, both of whom suffered impaired renal function before therapy began, high urinary levels of 5-fluorocytosine were obtained, which supports the findings of Koechlin *et al.* (1966) that this drug is mainly excreted in the urine. The fact that serum levels in this study were higher than those recorded by Shaddomy (1969) may be attributed to the fact that both patients suffered from partial renal failure, which may account for the persistence of the drug in the urine and serum after the last dose. Similar concentrations of 5-fluorocytosine have been recorded in serum by Record *et al.* (1971). No evidence of nephrotoxicity which might be attributed to the 5-fluorocytosine was observed. Though the drug was discontinued in Case 2 when the patient became marginally neutropenic, it cannot be concluded that this was the result of 5-fluorocytosine since his white blood cell count was variable before treatment. No platelet counts were undertaken.

In this study, though serum binding was estimated at 48–49%, no loss of inhibitory activity due to binding was observed in the experiments described. On the contrary, in low concentrations the fungistatic effect of 5-fluorocytosine on *T. glabrata* and *C. albicans* was enhanced as investigated by the addition of serum to Wickerham's medium (Table IV). A number of workers, including Hendry and Bakerspigel (1969), who reviewed the literature, reported on fungistasis in normal human serum. Chilgren *et al.* (1968) thought this was attributable to the clumping of cells in serum which was responsible for an apparent drop in the viable count with *C. albicans*. This is unlikely to have been the explanation in our study since clumps of cells were disrupted by ultrasonic vibration. Urine markedly inhibited the action of 5-fluorocytosine (Table III). Yet bioassay with *C. albicans* detected high levels of activity in diluted urine. It is possible that 5-fluorocytosine diffuses in agar at a different rate from the inhibitory substances which may occur in urine. Owing to the flushing of the bladder even a low level of fungistatic activity may suffice to eradicate infection in urinary candidiasis, and the first patient was undoubtedly cured of her urinary infection with *C. albicans*.

After using 5-fluorocytosine therapy for 15 patients with cryptococcosis, Utz *et al.* (1968) concluded that it was a promising agent because it was relatively non-toxic and had the advantage of oral administration. Their views are supported by Warner *et al.* (1970), who treated 10 patients with candidosis. Bennett (1970), however, reported that bone marrow or liver toxicity was seen in 3 out of 16 patients with

cryptococcosis and treated with from 4 to 6 g of 5-fluorocytosine daily. Webb *et al.*, (1970) described a *T. glabrata* septicæmia treated with 5-fluorocytosine and recorded that no toxic effects were observed during a short course with moderate blood levels. In view of these findings, at the present time 5-fluorocytosine appears to be the drug of choice for urinary yeast infections.

Since Shaddomy (1969, 1970) and Utz *et al.* (1968) reported resistance to 5-fluorocytosine in isolates of *Cryptococcus neoformans* and *C. albicans* from patients receiving the drug, sensitivity studies should be undertaken before and during therapy. A single step acquisition of resistance may occur and further laboratory investigation is required.

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